

In the process of evolution, animal and plant populations adapt to the conditions in which they live. When facing a climatic change there are several different ways for populations to react. They can tolerate the changed conditions as part of their phenotypic plasticity, they can shift their range in search of optimal conditions or adapt to new ones. However, if the changes are too profound or too fast, the populations may collapse. Understanding how populations respond to climate and environmental changes is therefore very important in the context of their protection, especially in view of the climate change ongoing currently. Responses of populations to climate changes in the past, such as demographic or adaptive changes, can be traced as a signal in the genomes of living individuals and hereby gained knowledge can help model and predict the responses of modern populations. However, inference from such data is quite limited. This is because similar patterns in the genome may arise from different processes and because consecutive processes often erase the signal of the previous ones. One of the possibilities to circumvent these limitations is the analysis of ancient DNA, i.e. DNA of individuals from palaeontological excavations. The analysis of the genomes of individuals from different periods gives a unique opportunity to directly track changes in the genetic diversity of the population and to reconstruct the processes that shaped it.

In the proposed project, we are planning to reconstruct the responses of a number of vole (*Microtus sp.*) species to the climate changes during the Late Glacial and Early Holocene using genomic sequences obtained from the fossil remains. The vole species are particularly suitable for such analyses. The remains of voles are abundant in palaeontological sites and that gives an opportunity to collect series of specimens covering a number of successive climate changes. The latest methods allow direct radiocarbon dating of even minute samples such as the vole remains, thanks to which it is possible to accurately put population changes on a timescale and compare their occurrence with the available palaeoclimate records. The PI's preliminary research also showed that vole molars contain well-preserved DNA in sufficient quantity and quality for analysis of nuclear genomes. However, due to DNA degradation, sequencing genomes from fossil remains is very difficult and expensive. Therefore, the first stage of the project will be to optimize a method that enables efficient genotyping of thousands of nuclear polymorphisms in multiple specimens.

We are planning to focus on three main themes. The first is the reconstruction of the post-glacial history of the population of common (*M. arvalis*) and field voles (*M. agrestis*) in the British Isles. The evolutionary history of both species has been extensively studied using mitochondrial DNA, but the colonization of the British Isles after the last Ice Age has not been fully resolved. In the case of common vole, its natural presence in the Isles has only recently been proven, and in the case of field vole, genetic data suggest that the Isles have been colonized at least twice and that natural selection was the main factor that shaped the modern population. We will obtain genomic data for several dozen fossil and modern individuals, which will make it possible to determine whether the British Isles were colonized once or several times, and what the role of natural selection was in the formation of today's populations.

The second issue will be the reconstruction and comparison of responses to climate change of four vole species in the Western Carpathians. For the comparison, we chose common vole, root vole (*M. oeconomus*), narrow-headed vole (*Lasiopodomys gregalis*) and bank voles (*Clethrionomys glareolus*). We are planning to obtain genome sequences for at least 50 specimens of each species. The selected species occupy a spectrum of ecological niches from cold and dry steppe-tundra to temperate forests. We expect that analysis of series of specimens from the Late Glacial and Early Holocene, periods which witnessed a series of profound climate and environmental changes, will reveal a variety of responses such as range shifts, adaptations, and local extinctions.

We are also planning to expand and refine the existing data on the environmental conditions prevailing in particular periods in the study area through metagenomic analyses of cave sediments. Such analyses complement reconstructions of plant cover based on pollen analyses, and the preliminary data obtained by the PI indicate that they can be successfully applied to cave sediments.

Combining the analysis of genomes of differently adapted species with accurate information on changes of environmental conditions over time will allow for robust determination the environmental factors that affected the studied species the most. The proposed project will allow not only to identify the population's responses to specific climate changes, it will also provide an opportunity to better understand the mechanisms of these responses.